By the present amendment, Applicant has amended Claims 26 and 46, and added new claims 47-52. Support for the new claims is found particularly on page 11, lines 5-8, and page 18, lines 15-22 of the specification. Claims 26, 28-43, and 45-52 are pending in the present application. Claim 26 is the sole independent claim. Applicant respectfully requests further examination and reconsideration of the application.

The Drawings Objection

Receipt of form PTO-948 is acknowledged. Applicant notes that the drawings as originally filed were objected to under 37 CFR 1.84. All informalities in the originally filed drawings will be corrected no later than upon submission of the issue fee.

The Sequence Disclosure Objection

Applicant has amended the specification to recite the appropriate SEQ ID NOs where sequences are disclosed in the specification.

The Section 112 Rejections

Claims 26, 28, 45, and 46 were rejected under 35 USC 112, second paragraph, as being indefinite. The Examiner objected to the expression "essentially preserve the overall tertiary structure" in independent Claim 26 as unclear. This rejection is respectfully traversed. The specification does not give any *expressis verbis* definition of this phrase. However, the specification does disclose a teaching which would guide the person skilled in the art to a precise understanding of the meaning of "essentially preserve the overall tertiary structure".

In the Description of the Prior Art, specification pages 2-5, various prior art techniques are discussed which are designed to raise immune responses against self-proteins. One of these is traditional conjugation technology. The specification describes a drawback of this technology, in that "...the antibody response towards the self-protein will be limited due to shielding of epitopes by the covalently linked carrier protein..." (page 2, lines 29-31). This implies that the modified self-proteins of Applicant's invention do not suffer this drawback of "losing" a number of B-cell epitopes. This must further have the consequence that essential preservation of overall tertiary structure results in the conservation of substantially all B-cell epitopes of the self-protein. This interpretation of "essentially preserve the overall tertiary structure" is in accordance with the discussion on page 4, lines 7-10, which emphasizes that the tertiary structure determines the specific recognition of the non-modified self-protein by the induced antibodies. See also page 4, lines 27-29. Specific functional changes are therefore what is referred to, not changes to the crystal structure. See page 8, lines 27-31.

Page 8, line 33 states that the tertiary structural changes are minimal. The skilled person in protein chemistry will readily be able to identify regions in a given protein which will be suitable for the introduction of a foreign T-cell epitope by means of substitution while ensuring the claimed preservation of overall tertiary structure. Likewise, for a third party who attempts to induce antibody production in an animal, it will be easy to determine whether the immunogen used contains an in-substituted T-cell epitope and has preserved the overall tertiary structure (*i.e.* has preserved substantially all B-cell epitopes of the native protein).

The Examiner also objected to the term "flanking regions" in Claim 46 as unclear. Claim 46 has been amended to replace the term "flanking regions" with "amino acid sequences".

Applicant respectfully submits that Claims 26 and 46, as well as the other pending claims, comply with the requirements of 35 USC 112.

The Prior Art Rejections

The Examiner rejected Claims 26, 28, 45, and 46 under 35 USC 102(b) as being anticipated by Russell-Jones et al., WO 92/05192. For a prior art reference to anticipate a claimed invention, the prior art reference alone should either explicitly or implicitly disclose each and every aspect of the claimed invention, MPEP 706.02. The Russell-Jones reference fails to disclose at least three elements of Applicant's claimed invention: The use of a modified self-protein as an immunogen, substitution in the self-protein sequence, and preservation of the overall tertiary structure of the unmodified self-protein. The disclosure of Russell-Jones is also insufficient to render Applicant's claims obvious.

Note that independent Claim 26 has been amended to clarify that a single fragment of the self protein may be substituted by a T-cell epitope. Claim 26 has also been amended to remove a minor inconsistency (use of the term "animal" instead of "animal species").

**Modified self-proteins vs. heterologous protein analogues**

Independent Claim 26, on which all of Applicant's pending claims are dependent, includes the step of administering a self-protein analogue, or "modified self-protein", to an animal. The modified self-protein is modified from the amino acid sequence of the unmodified self-protein by containing a substitution of a peptide fragment of the self-protein with a peptide containing a foreign, immunodominant T-cell epitope. The modification of the self-protein is limited in that the overall tertiary structure of the unmodified self-protein is preserved.

The goal of administering Applicant's modified self-protein is the breaking of the animal's immune tolerance against the self-protein. A variety of means of breaking autotolerance to self-proteins have been reported in the prior art. See specification, pages 2-5. A common approach to breaking autotolerance has been to administer heterologous protein in the form of a homologue of the self-protein derived from a different animal species. See specification, pages 2-5, and enclosed prior art abstracts. For example, see the attached abstract of Talwar GP et al., Fertil. Steril. 46: 120-126 (1986), which teaches the immunization of monkeys with ovine LH beta subunit coupled to tetanus toxoid as carrier.

Two abstracts published after the priority date of the present application are also attached, to show that this approach is still contemplated seriously. The abstract of Tannetta DS et al., J. Reprod. Fertil. 110: 255-262 (1997), discloses the immunization of ewes with bovine inhibin a1-29 conjugate. The abstract of Kaliyaperumal A et al., Eur. J. Immunol. 25: 3375-3380 (1995) teaches the immunization of women with the $\alpha$-chain of ovine LH coupled to diphtheria toxoid and tetanus toxoid, and notes that a change of carriers has a significant effect.

As shown by the attached abstracts, known prior art methods involve administration of a poorly immunogenic, heterologous protein which shares sufficient homology with the target self-protein for down-regulation. In experimental animals it has been necessary to use strong adjuvants or foreign carrier proteins to induce a satisfactory immune response. For example, the Talwar abstract indicates that since CFA is unacceptable for human use, tetanus toxoid was tested instead.

The Examiner states that the Russell-Jones reference teaches that the immunogens used in the disclosed vaccines include self proteins such as LH, somatostatin, inhibin, and FSH. Russell-

Jones does not mention that these are self-proteins, and provides no guidance as to the specific origin of these hormones. The Russell-Jones reference is concerned with *improving* the immune response against immunogens, while the present invention is concerned with rendering *non-immunogenic* self-proteins immunogenic. The skilled person would interpret the teaching in Russell-Jones as an indication that the TraT epitopes can be used, instead of known carrier proteins, to render *heterologous* LH, FSH, somatostatin and inhibin more immunogenic.

Since Russell-Jones mentions nothing relating to immunization against self-proteins, the skilled artisan would be required to turn to the prior art. Such a search, as stated above, would merely prompt the use of TraT peptides in a vaccine utilizing heterologous versions of LH, FSH, somatostatin and inhibin.

Note that Russell-Jones uses the term "immunogen"; this term cannot be used meaningfully with regard to non-immunogenic self-proteins. See Abstract and page 8, lines 19-25 and 36-38. Russell-Jones states that poorly immunogenic immunogens are preferred, page 9, lines 1-3. Heterologous versions of LH, FSH, somatostatin, and inhibin are poorly immunogenic, as Russell-Jones indicates on page 9, lines 1-12. However, the autologous versions of these proteins are not poorly immunogenic; rather they are not immunogenic at all. Russell-Jones provides no incentive to use TraT peptides or other short peptides to break autotolerance to a self-protein. Russell-Jones does not teach or suggest that the immunogens are derived from the animal which is going to be immunized. Since the prior art also does not suggest this, it must be concluded that the immunization suggested in Russell-Jones follows the normal prior art route of immunization with heterogeneous material.

**Substitution vs. insertion**

The Examiner relies on Russell-Jones to show insertion of TraT peptides into the immunogen, via substituting TraT peptide for a peptide contained in the immunogen. The use of substitution, rather than insertion, is an essential feature of the present invention. Applicant's specification makes clear that a substitution is a combination of the two basic operations of deletion and insertion, page 8, lines 27-31, and page 10, lines 7-13. The goal of Applicant's technology is to produce immunogens which preserve a maximum number of B-cell epitopes of an animal's self-protein to allow for induction of a diverse antibody response. It is therefore necessary to ensure that the resulting modified self-protein resembles the native protein to the highest possible degree — hence the requirement of preserving the overall tertiary structure of the self-protein. Substitution ensures that the length of the modified protein will not be substantially different from that of the native protein, optimizing the chances of preserving overall tertiary structure.

The text referred to by the Examiner on pages 30 and 31 of Russell-Jones relates to *insertion* of TraT epitopes, not to substitution of part of an antigen with these epitopes. See page 31, lines 4-8. The suggested use of LH as an immunogen involves insertion of LH in TraT or in a part thereof to place LH adjacent to a TraT T-cell epitope. See page 9, lines 20-26. This is not in any way the same as substitution of a T-cell epitope into a self-protein.

The only location in which Russell-Jones suggests substitution is the Examiner's cited text on page 32. However, the antigen involved in this substitution is a *viral* (HIV) protein, page 32, lines 3 and 25. The immunogen is clearly not a self-protein of an animal. The gp120/gp41 complex is immunogenic in humans and gives rise to antibody production in vivo, whereas self-

proteins are totally non-immunogenic in most individuals. Further, the rationale behind the disclosed substitution is to remove "suppressor regions" which interfere with the development of effective immune responses to the antigen having these suppressor regions. Nothing in Russell-Jones suggests that suppressor regions exist in any self-proteins, much less in FSH, somatostatin, LH, and inhibin. Note that Example 5 reports no results showing that this strategy has in fact proven effective in producing an antibody response, even against HIV. Applicant is not aware of any successful later attempts to immunize against HIV using the strategy of Example 5, which suggests that Russell-Jones may not be enabling for induction of an improved anti-HIV antibody response. The skilled person would not conclude from the disclosure of Russell-Jones that the specific strategy of Example 5 would be generally applicable, let alone applicable for rendering non-immunogenic self-proteins immunogenic.

**Preservation of overall tertiary structure**

The Examiner relies on Russell-Jones to show that the TraT peptide is inserted into the immunogen so as to essentially preserve the overall tertiary structure. However, page 32 of Russell-Jones, which is the only location in which substitution is mentioned, does not suggest that such substitution preserves the overall tertiary structure of the unmodified protein. On the contrary, Example 5 suggests the removal of a suppressor region in gp41 (SEQ ID NO: 18 of Russell-Jones, page 32, lines 25-30) which together with gp 120 forms a complex protruding from the outer envelope membrane of HIV. The transmembrane protein gp41 has a length of about 350 amino acids, with the exact length and sequence depending on the HIV-1 isolate. (Note that while Russell-Jones refers consistently to suppressor regions of gp120, the amino acid sequence given in Example 5 is present in gp41 and not in gp120.) Substitution of the suggested

suppressor region would be very likely to cause a significant change in tertiary structure, particularly as no precaution against it is mentioned. As discussed above, in Applicant's claims essential preservation of overall tertiary structure results in the conservation of substantially all B-cell epitopes of the self-protein.

The Examiner indicates that tertiary structure is maintained because the ability of the immunogen to function as an immunogen is maintained. As discussed above, it is likely that immunogenicity is not in fact maintained using the strategy of Example 5 of Russell-Jones, given the lack of later success using this approach in enhancing immunogenicity of the HIV protein. Certainly no motivation would be found to use this strategy with a self-protein instead of a viral protein.

In addition, maintenance of immunogenicity is neither required nor sufficient for essential preservation of overall tertiary structure as required by Applicant's claims. Russell-Jones is concerned with immunogenic non-self-proteins. Accordingly, it is sufficient to preserve single immunogenic epitopes. Applicant's self-proteins are non-immunogenic in the host before the T-cell epitope is inserted. Applicant's claims require the conservation of substantially all B-cell epitopes of the self-protein, so that any change in tertiary structure must be minimal. The overall tertiary structure of a protein cannot be said to be preserved if a single immunogenic epitope is isolated, and its immunogenic character is preserved by coupling to a carrier.

The example of somatostatin in Russell-Jones, mentioned by the Examiner, if anything, teaches away from the maintenance of overall tertiary structure. See page 9, lines 9-12. Biologically active somatostatin is a peptide having a length of only 14 amino acids, which is derived from a 28 amino acid precursor. The insertion of a TraT peptide of even 14 amino acids

(which is the shortest TraT peptide disclosed in Russell-Jones; see SEQ ID NO: 2) would certainly destroy most, if not all, tertiary structure of somatostatin.

**Newly introduced claims**

New Claims 47-52 relate to the method of the invention in which the modified self-proteins are specified as being modified TNF-α, TNF-β, γ-interferon, IL-1 or IgE. None of these self-proteins are mentioned or suggested in Russell-Jones. With respect to TNF-α, Example 3 demonstrates that the vaccination method provides surprising results. In particular, the antibody response against TNF-α was not restricted to the MHC classes recognizing the implanted T-cell epitopes. See specification, page 13, lines 23-30. Further, it is demonstrated in Example 4 that the method of the invention is superior compared to traditional conjugation technology, page 14, lines 16-20.

For at least these reasons, Applicant respectfully submits that independent Claim 26, as well as all the dependent claims, is allowable over the references cited by the Examiner. Reconsideration and withdrawal of the grounds of rejection are respectfully requested.

Summary

Applicant respectfully submits that the present application is in condition for allowance. In the event there are any issues which can be expedited by telephone conference, the Examiner is cordially invited to call the undersigned at the number indicated below. Authorization is hereby granted to charge payment of any additional fee to Deposit Account 06-1358.

Respectfully submitted,

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Enclosures: Petition for Extension of Time
IDS with PTO-1449
Prior art abstracts: Talwar, Al-Obaidi, O'Shea, Thau, Thau, Moudgal, Wickings
Other abstracts: Kaliyaperumal, Tannetta

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